Respiratory Medicine (2012) 106, 1096–1102

Available online at www.sciencedirect.com

SciVerse ScienceDirect

journal homepage: www.elsevier.com/locate/rmed

The use of rhDNAse in severely ill, non-intubated adult asthmatics refractory to bronchodilators: A pilot study☆

Robert A. Silverman [b,*], Finbar Foley [a,d], Resul Dalipi [a,e], Myriam Kline [b,c], Martin Lesser [b,c]

[a] *Department of Emergency Medicine, Long Island Jewish Medical Center, North Shore-LIJ Health System, NY 11040, USA*
[b] *Feinstein Institute for Medical Research, North Shore-LIJ Health System, Manhasset, New York, USA*
[c] *Department of Biostatistics, Feinstein Institute for Medical Research, North Shore-LIJ Health System, Manhasset, New York, USA*

Received 21 June 2011; accepted 4 April 2012
Available online 12 May 2012

KEYWORDS
Asthma;
Acute;
Severe;
rhDNAse

Summary
Introduction: Mucous plugging is associated with fatal asthma and may have a causative role for non-fatal cases of severe acute asthma. However, mucolytic agents have not been found effective in reversing the obstruction of acute asthma. We test the hypothesis that rhDNAse, an agent that reduces viscoelasticity of sputum in patients with cystic fibrosis, has a therapeutic role in acute asthma.
Methods: Symptomatic asthmatics aged 18–55 years presenting to an Emergency Department with an $FEV_1 < 60\%$ predicted after 2 nebulized albuterol and ipratropium treatments were included. Patients were randomized into one of three nebulized rhDNAse treatment groups of2.5, 5.0 or 7.5 mg, or placebo. Standardized bronchodilator therapy was continued throughout the protocol and the FEV_1 at 6 h was the primary study endpoint.
Results: 50 patients were enrolled. There were no significant differences in FEV_1% predicted between the rhDNAse and placebo patients at any of the post-randomization time points. The dose of rhDNAse administered did not influence response. In a post-hoc stratification, patients with the lowest pre-randomization FEV_1 tended to improve more from rhDNAse,

Abbreviations: CI, Confidence Interval; ED, Emergency Department; FEV_1, Forced expiratory volume in the first second of exhalation.

☆ Study site: Emergency Department of the Long Island Jewish Medical Center.

* Corresponding author. Department of Emergency Medicine, Long Island Jewish Medical Center, North Shore-LIJ Medical Center, New Hyde Park, NY 11040, USA. Tel.: +1 718 470 7501; fax: +1 718 470 9113.
E-mail address: aresilv@aol.com (R.A. Silverman).
[d] Present address: Johns Hopkins School of Medicine, Baltimore, MD, USA.
[e] Present address: Stamford Health System, Columbia University College of Physicians and Surgeons, Stamford, CT, Milford Hospital, Milford, CT, USA.

doi:10.1016/j.rmed.2012.04.002

particularly at times 60 and 120 min post-randomization.
Conclusion: In this pilot study rhDNAse did not cause clinical improvement among severely ill adults refractory to standardized care. The observed trend to higher FEV_1 among the most severely obstructed patients is an exploratory finding that may warrant further study.
This clinical trial was registered as NCT00169962 under the name "Study of Pulmozyme to Treat Severe Asthma Episodes".

Introduction

Airway obstruction in severe acute asthma can be caused by bronchoconstriction, airway edema, and mucous plugging. Bronchoconstriction is typically treated with short acting beta-agonists, inhaled anticholinergics and when severe, intravenous magnesium sulfate.[1,2] Systemic steroids are used to treat airway edema and inflammation, although benefit in adults is not always noted during the Emergency Department (ED) treatment period. In spite of bronchodilator and prednisone based ED therapy, of the 1.8 million annual ED visits for asthma in the USA nearly 500,000 hospitalizations still occur.[3] This is in addition to 4200 annual deaths attributed to asthma.[3]

Fatal asthma is typically characterized by exudative airway luminal obstruction with extensive plugging that is very difficult to reverse.[4] Presumably patients with severe acute asthma who do not respond to bronchodilators are more likely to have acute and subacute mucous plugging contributing to airway obstruction. The pathologic findings are consistent with the observation that shorter durations of asthma worsening respond more quickly to bronchodilators than longer durations.[5]

Mucoactive agents to enhance clearance of mucin-related secretions have been investigated for acute severe asthma without clear evidence of efficacy, and NAEPP asthma guidelines do not recommend mucolytics to reverse airway plugging.[6] Aerosolized rhDNAse is an FDA approved agent for cystic fibrosis used to alleviate airway obstruction from viscous secretions.[7] Several anecdotal reports have found rhDNAse effective in reversing mucous plugging in either intubated patients or non-intubated patients refractory to beta-agonists.[8–10] However, in the only published clinical trial evaluating the efficacy of rhDNAse, acutely ill asthmatic children with moderate to severe exacerbations did not show benefit.[11]

Our objective was to determine if aerosolized rhDNAse was a useful adjunct in non-intubated adult ED asthmatics with very severe acute obstruction refractory to nebulized bronchodilators. We evaluated 1) whether rhDNAse improved pulmonary function in acute severe asthma 2) whether increasing doses of rhDNAse provided greater improvement in pulmonary function, 3) the relationship between illness severity and response to rhDNAse and 4) safety of rhDNAse in acute asthma.

Methods

The study was conducted in the Emergency Department (ED) at Long Island Jewish Medical Center, a voluntary teaching hospital. Inclusion criteria were patients ages 18–55 years with acute asthma worsening, a 6 month or longer history of known asthma, and a forced expiratory volume in 1 s (FEV_1) less than 60% of predicted at both ED entry and 40 min after two sets of standardized asthma treatments (see trial design below). Patients with the following were excluded: greater than 15 pack-year history of smoking; pregnant or breast feeding; a need for intubation before randomization; pneumonia; chronic lung disease other than asthma; any other clinically significant medical conditions that could in the opinion of the investigator increase the risk of adverse effects with treatment. Additionally, patients had to be willing and able to perform spirometry maneuvers, stay in the ED for at least 6 h, and then participate in a follow-up visit in 24 h.

Trial design and treatment

Upon ED arrival patients underwent spirometry and a brief clinical assessment followed by treatment with 5.0 mg nebulized albuterol, 0.5 mg nebulized ipratropium, and either 60 mg oral prednisone or 125 mg intravenous methylprednisilone. For patients with an initial $FEV_1 < 25\%$ predicted 2 g of magnesium sulfate was infused intravenously over 10 min. After the initial bronchodilator treatment was completed (approximately 20 min after ED entry), spirometry was repeated and a second course of 5.0 mg nebulized albuterol and 0.5 mg nebulized ipratropium given. After completion of this second bronchodilator treatment spirometery was repeated and patients with an $FEV_1 < 60\%$ who met study inclusion criteria were randomized to receive study interventions or placebo.

This was a dose ranging study where a total of 50 patients were randomized 3:2 to receive double-blind, single-dose treatment with 2.5 mg of nebulized rhDNAse or placebo (first 17 patients); 5.0 mg nebulized rhDNAse or placebo (next 17 patients), or 7.5 mg nebulized rhDNAse or placebo (final 16 patients) (Fig. 1). Therefore a total of 30 patients received a single dose of rhDNAse and 20 patients received a like quantity of placebo sterile saline. After study drug administration the standardized treatment protocol continued, with patients receiving 2.5 mg albuterol and 0.5 mg ipratropium at time 40 min; additional 2.5 mg albuterol treatments were administered at time 60 min and hourly until 360 min after ED arrival. All inhaled treatments were given by a disposable hand held nebulizer driven by a 7 L 100% oxygen flow, and administered directly by mouthpiece. Over the 360 min protocol each patient received a total of 22.5 mg of albuterol and 1.5 mg of ipratropium.

At the 360 min ED assessment (approximately 320 min after trial drug administration), the investigator evaluated

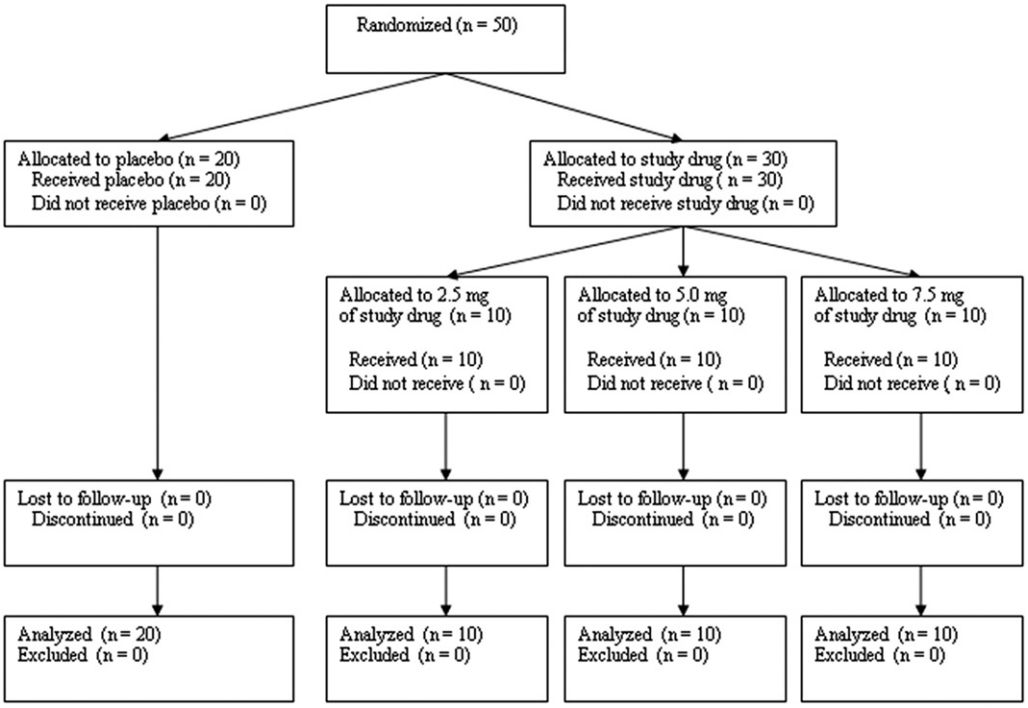

Figure 1 CONSORT diagram of enrolled patients.

whether patients could be discharged or required hospital admission. Clinicians were asked to adhere to NAEPP consensus guidelines for determining need for hospitalization, and to further provide consistency the protocol recommended hospitalization for a final $FEV_1 < 50\%$ predicted, although the treating clinician had the final disposition decision. 24 h after enrollment, patients had repeat clinical assessments which included spirometry and recording of any adverse events. A follow-up telephone contact was made 7 days after trial treatment ended, and patients described any additional urgent or emergent need for medical care since the 24 h assessment.

rhDNAse and placebo nebules were identical in appearance and equal in volume, and both the patient and investigator were blinded to the intervention. Randomization schemes were computer generated by the hospital pharmacy. Written informed consent was obtained from all patients and the study was approved by the investigational review board of the North Shore-LIJ Medical Center.

Spirometry methods were based on American Thoracic Society/European Respiratory Society criteria with modifications made for a trial population of acutely ill subjects.[12,13] Spirometry was performed using the KoKo Spirometer (Pulmonary Data Services, Lewisville, Colo), with real-time feedback provided to ensure compliance with study performance criteria. Measured value-acceptability criteria for patient maneuvers included a forced expiratory effort of at least 2 s, a back-extrapolated volume of $<5\%$, and a peak flow rate within the first 120 ms of forced expiration. Three or more efforts were obtained at each assessment. FEV_1 values were considered reproducible if the two best efforts were within 10% of each other. An additional criteria, absence of glottic closure in the first second, was evaluated when time—flow graphs were inspected visually. Other clinical assessments included peak flow rate, respiratory rate, use of accessory muscle usage (sternocleidomastoid muscle groups, yes/no) and a modified Borg scaled dyspnea index which utilized a numeric scoring system (10 = most severe dyspnea, 0 = none) along with visual cues.

Statistical analysis

This study was designed to explore the efficacy, tolerability and safety of rhDNAse in an acutely ill ED population. $FEV_1\%$ predicted at 6 h after ED arrival was the primary endpoint. In addition we performed a number of planned exploratory analyses to test several hypotheses. This included determining whether rhDNAse improved FEV_1 at a number of post-randomization time points; determining whether higher rhDNAse doses led to greater improvement; and determining whether patients with the lowest FEV_1 were more likely to respond to rhDNAse. Since the study was set up as exploratory, power analyses were not used to determine the sample size.

Unadjusted Student's *t*-tests were used to compare FEV_1 % predicted for the primary 360 min hour endpoint and for each of the post-randomization time points(time 60,120, 180, 240 and 360 min after ED arrival). The post-randomization FEV_1 % predicted data were then adjusted for the pre-randomization FEV_1 (time 40 min) to control for variability in the initial measurement. To test whether severity of illness influenced the drug effect, patients were split into 2 groups according to the pre-randomization (time 40 min) median FEV_1 % predicted value. The model included treatment group (drug), time 40 min FEV_1 % predicted (severity), and the interaction between drug and severity (drug*severity) for each of the time points.

Results

Fifty patients were enrolled and clinical characteristics are described in Table 1. The placebo and rhDNAse subjects were similar for age, gender and past asthma history as well as FEV_1 % predicted on ED arrival. Other clinical parameters appeared similar for the rhDNAse and standard care groups on ED arrival, including peak flow (38.9% versus 39.2% predicted), minute respiratory rate (21.9 versus 20.0) and frequency of accessory muscle/sternocleidomastoid usage (79% versus 65%). Both groups had a similar response to the pre-randomization bronchodilator treatments given at time 0 and 20 min, and at time 40 min the FEV_1 was 38.4% predicted in the rhDNAse group and 39.0% predicted in the standard care group.

Table 2 presents the FEV_1 % predicted for the placebo and rhDNAse groups at all study time points. The FEV_1 at 360 min was similar in the placebo and rhDNAse groups ($p > 0.05$) as well as for all other post-randomization time points. Data was then adjusted for the pre-intervention FEV_1 (time 40 min) and shown in Fig. 2. Again, there were no differences in FEV_1 between placebo and rhDNAse groups at any of the post-randomization time points($p > 0.05$). Subjective dyspnea symptoms were assessed by a modified Borg scale, with 10 representing the most severe dyspnea and 0 representing no dyspnea symptoms. There were no significant differences between the 2 groups at any of the post-randomization points (data not shown), and at time 360 min the Borg score in the placebo group was 2.7 and in the rhDNAse group was 3.4 (p = ns), There weren't any significant differences in heart or respiratory rates at any of the post-randomization time points (data not shown).

Table 1 Patient characteristics at ED presentation.

	rhDNAse ($N = 30$)	placebo ($N = 20$)
Demographic characteristics:		
Age in years, mean (SD)	41.3 (9.7)	36.0 (10.2)
Female, *n* (%)	21 (70%)	15 (75%)
Race/Ethnicity, *n* (%)		
White	4 (13.3%)	1 (5%)
Black	18 (60%)	9 (45%)
Asian	2 (6.7%)	1 (5%)
Hispanic	3 (10%)	6 (30%)
Other	3 (10%)	3 (15%)
Asthma history:		
ED visits past 1 yr (median,range)	1.0 (0—5)	1.0 (0—10)
Hospitalizations past 5 yrs (median,range)	0.0 (0—6)	0.0 (0—12)
Previous intubations, *n* (%)	6 (20%)	4 (20%)
ICU asthma visits past 5 yrs, *n* (%)	0 (0%)	1 (5%)
Inhaled steroid usage past 6 months, *n* (%)	8 (30.8%)	5 (35.7%)
Smoker (current or previous), *n* (%)	11 (36.7%)	13 (65.0%)
No. of yrs with asthma, mean (SD)	22.8 (14.9)	20.7 (14.2)

Table 2 Average FEV_1% predicted (unadjusted).

Time (minutes)	rhDNAse FEV_1% predicted($N = 30$)	placebo FEV_1 % predicted ($N = 20$)
0	28.2	30.1
20	35.1	36.5
40	38.4	39.0
60	40.9	42.7
120	43.6	45.6
180	47.0	45.7
240	47.9	45.8
360	47.0	47.5
24 HR	39.6	43.8

rhDNAse or placebo was administered at time 40 min. Comparing FEV_1 at each time point, all p values > 0.05.

To explore whether more severe obstruction predicted response to therapy, Table 3 shows data grouped by the pre-randomization (time 40 min) FEV_1 above and below 39% (the median value). Patients with a $FEV_1 \leq 39\%$ predicted had responses in favor of the drug at time 60 min ($p = 0.045$) and 120 min ($p = 0.039$). The time 180, 240 and 360 min p values were greater than 0.05 at these later time points. When adjustment for multiple comparisons were performed, significance was lost at all time points.

Fig. 3 indicates the differences in FEV_1 at the post-intervention time points for each of the three rhDNAse doses when compared to the placebo group. There was no clear pattern of a dose-related effect. A regression analysis using the three different doses of rhDNAse and adjusting for the $t40FEV_1$ did not find any apparent dose-related effect on pulmonary function (data not shown).

Hospitalization and follow-up

The hospitalization rates in the placebo group (45%) and in the rhDNAse group (63%) did not differ significantly ($p > 0.05$). After adjustment for FEV_1 at time 40 min,

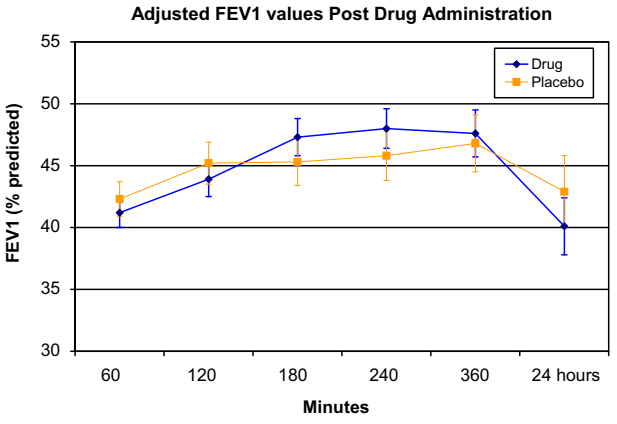

Figure 2 Change in FEV_1 over 6 h treatment period and 24 h follow-up for both investigational product and placebo. FEV_1 data was adjusted for pre-intervention values. Error bars indicate the standard error of the mean.

Table 3 Percent predicted FEV_1 for post-ED arrival times by rhDNAse use and severity at time 40 min post-ED arrival*.

Minutes post-ED arrival	$FEV_1 > 39\%$ predicted		$FEV_1 \leq 39\%$ predicted		Interaction between drug and severity *p*-value
	placebo $N = 12$	rhDNAse $N = 13$	placebo $n = 8$	rhDNAse $N = 17$	
Time 60	52.0 ± 9.3	49.1 ± 8.1	28.8 ± 4.9	34.7 ± 6.0	0.045
Time 120	55.5 ± 12.5	51.2 ± 8.7	30.8 ± 6.3	37.8 ± 7.6	0.039
Time 180	54.9 ± 12.7	54.8 ± 10.2	31.9 ± 6.9	41.0 ± 8.8	ns
Time 240	54.8 ± 11.9	55.8 ± 11.3	33.5 ± 6.9	42.4 ± 9.1	ns
Time 360	56.8 ± 13.9	54.0 ± 10.8	33.9 ± 8.6	42.5 ± 10.1	ns
Time 24 h	50.0 ± 13.7	42.5 ± 12.8	32.5 ± 8.8	37.6 ± 12.7	ns

*Data was partitioned and analyzed into two groups based on the median FEV_1 at time 40 min (pre-randomization value). Please see text for details.

hospitalization risk for patients receiving rhDNAse did not differ significantly from placebo patients (OR 2.16 (95% CI 0.63—7.34)). Time 24 h spirometry measures were obtained in 44/50 patients. After adjustment for FEV_1 at time 40, the mean FEV_1 in the placebo group was 42.9% and in the rhDNAse group was 40.1% ($p > 0.05$). In addition, there were no differences between the groups for relapse at day 7.

Safety

There were 2/30 patients in the rhDNAse group and 0/20 in the placebo group with more than a 10% decrease in FEV_1 at the first study assessment point following the study intervention (time 60 min). In one patient who received 2.5 mg rhDNAse the FEV_1 changed from 36% predicted at time 40 min to 31% at 60 min; the time 360 min FEV_1 was 39% predicted. The FEV_1 in another patient who received 5.0 mg rhDNAse went from 60% predicted at time 40 min to 36% at 60 min. This patient had a past history of multiple intubations and after study completion she indicated prior episodes of severe bronchospasm after receiving any type of nebulized therapy. She continued in the study and by time 120 min her FEV_1 was 52% predicted and at 240 min FEV_1 was 59% predicted. There were no other clinically significant adverse effects noted in other patients during the study.

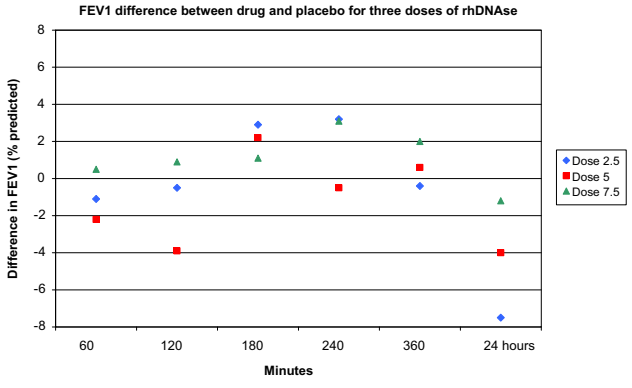

Figure 3 Difference of means (drug minus placebo) for each dosing level of investigational product compared to placebo group over the 6 h treatment period and for the 24 h follow-up. Data was adjusted for pre-randomization values.

Discussion

We explored whether a single dose of nebulized rhDNAse improved pulmonary function in non-intubated severely obstructed Emergency Department asthmatics refractory to standardized bronchodilator therapy. Overall rhDNAse did not cause significant improvement in FEV_1 at any of the post-randomization time points. rhDNAse did not have an impact on need for hospitalization or on the FEV_1 24 h after randomization.

When we evaluated data from patients with more severe obstruction, those receiving rhDNAse tended to have more improvement in pulmonary function especially at the earlier time points, but small sample size and multiple exploratory analyses limit interpretation of these findings.

There are a number of published case reports of patients with life-threatening asthma who were failing conventional management and responded to rhDNAse either by direct intratracheal instillation[8]; direct instillation of rhDNAse via bronchoscope[10,14]; and in a case of an intubated pregnant asthmatic, nebulization via the endotracheal tube.[15] In a case-series of 8 intubated asthmatic children, intratracheal rhDNAse followed by percussive physiotherapy improved ventilation.[16] Three other cases of non-intubated children responding to nebulized rhDNAse have also been reported.[9]

One previous clinical trial evaluating rhDNAse has been published.[11] Among 121 children with acute moderate to severe asthma, 5.0 mg of nebulized rhDNAse did not improve asthma scores or the need for hospitalization. There was a slightly better trend to improved asthma scores in the rhDNAse group at the 60 and 120 min post-randomization time points; this was not statistically significant. There were no previous published clinical trials evaluating rhDNAse in adults.

The rationale for attempting to reverse mucous plugging in severe acute asthma is supported by autopsy studies. Extensive airway obstruction by exudate containing both mucus and cells is characteristic of fatal asthma episodes[4,17] with plugging involving large and small airways.[18] Airway exudates are composed of mucin, plasma proteins, DNA and a mixture of inflammatory and epithelial cells. Cellular material has been found to be as important as mucus and protein exudates in luminal occlusion of fatal obstructive asthma,[4] especially in smaller airways.

In designing this pilot study we postulated mucous plugging was an important factor in severe airway compromise, and that patients with the most extensive mucous plugging would benefit the most from rhDNAse. Since we had no direct means to identify the extent of mucous plugging in an ED setting, the lack of response to bronchodilator therapy was used as an indicator of airway plugging. This provided a practical means of testing our hypothesis in non-intubated patients, but did not provide certainty regarding the contribution of plugging to the acute episode and this remains a study limitation. Future studies may consider using 24 h response to systemic steroids and continued usage of bronchodilator therapy as an alternative method of selecting patients with presumed mucous plugging.

Factors potentially influencing the efficacy of rhDNAse include the content of DNA relative to other substances (eg mucin) that contribute to the viscosity of the plug, and the size and location of the mucous plug. We did not know whether the amount of DNA contained in the mucous of patients with severe asthma was sufficient to allow reversal of mucous plugging after treatment with rhDNAse. Sputum from asthmatics have higher DNA content than sputum from non-asthmatics.[19] While the DNA content is much higher in pulmonary secretions from cystic fibrosis patients compared to patients with chronic stable asthma, it is possible patients with acute but non-fatal asthma would have higher sputum DNA content than individuals with chronic stable asthma. However, we did not find any published studies which evaluated DNA content of airway plugs in patients surviving a severe episode.

We also assumed (but had no way to prove) enough nebulized drug would reach occluded airways to promote expectoration of the plug. Presumably much higher amounts of drug reach the sites of plugging when installed via the intratracheal route or bronchoscopic visualization, and more plug could be removed when mechanical suction is applied compared to the cough mechanism. This study limitation can be addressed in future studies by testing repeated dosing strategies in the most severely ill non-intubated patients, or through a trial involving intubated patients.

Regarding safety, in two patients a 10% drop in FEV_1 occurred immediately following administration of rhDNAse which subsequently increased with beta-agonist therapy. Since most participants, including those with more severe obstruction, had improvement soon after receiving rhDNAse we cannot determine whether the drop in FEV_1 was incidental or whether we identified a subgroup of patients with drug induced bronchoconstriction. In addition, we noted a non-significant trend for increased hospitalization and lower FEV_1 at 24 h in patients who received rhDNAse. One possible explanation is patients subsequently randomized to rhDNAse had a lower FEV_1 on ED arrival (before administration of rhDNAse), and they reverted back to the lower baseline after completion of aggressive ED bronchodilator therapy. Further study is needed to determine whether these findings are due to chance randomization or the drug itself.

In summary, rhDNAse did not improve airway obstruction in non-intubated ED patients with severe asthma refractory to bronchodilators. Slightly greater improvement in FEV_1 was found among patients with lower initial FEV_1 but small sample size and multiple analyses limit interpretation of these findings. This study does not support routine usage of rhDNAse in patients with very severe acute asthma, but rather identifies a subgroup of patients where the use of rhDNAse could be further studied.

Funding

This work was supported by an unrestricted investigator initiated research grant awarded by Genentech to Robert Silverman MD.

Conflict of interest statement

Robert Silverman has received funding from AstraZeneca, Medimmune and MediciNova.

Finbar Foley has no conflicts of interest to disclose.

Rasul Dilipi has no conflicts of interest to disclose.

Myriam Kline has no conflicts of interest to disclose.

Martin Lesser has no conflicts of interest to disclose.

Acknowledgments

Robert Silverman wrote the protocol, obtained funding, supervised data collection, participated in the data analysis and drafted the manuscript. Finbar Foley and Resul Dalipi helped in the development of the protocol and data collection. Martin Lesser, Myriam Kline and Edith Flaster conducted the statistical analyses.

We also acknowledge Ray Pahk, Rosetta Grella, Andrew Green and Michelle Carbone for assisting with data collection, and Reed Magleby for assistance with manuscript preparation.

ClinicalTrials.gov identifier: NCT00169962.

References

1. Silverman R, Osborne H, Runge J, et al. IV magnesium sulfate in the treatment of acute severe asthma: a multi-center randomized controlled trial. *Chest* 2002;**122**:489—97.
2. Rowe BH, Bretzlaff JA, Bourdon C, Bota GW, et al. Magnesium sulfate for treating exacerbations of acute asthma in the emergency department. *Cochrane Database Syst Rev* 2000;**1**: CD001490.
3. Moorman JE, Rudd RA, Johnson CA, et al. National Surveillance for asthma —- United States, 1980—2004. *MMWR CDC Surveill Summ* 2007;**56**(SS08):1—14. 18-54.
4. Kuyper LM, Pare PD, Hogg JC, et al. Characterization of airway plugging in fatal asthma. *Am J Med* 2003;**115**:6—11.
5. Woodruff PG, Edmond SD, Singh AK, et al. Sudden onset severe acute asthma: clinical features and response to therapy. *Acad Emerg Med* 1998;**5**:695—701.
6. National Asthma Education and Prevention Program. *Expert Panel report 3: guidelines for the diagnosis and management of Asthma*. Bethesda, MD: U.S. Department of Health and human Services, National Institute of Health; 2007.
7. Fuchs HJ, Borowitz DS, Christiansen DH, et al. Effect of aerosolized recombinant human DNase on exacerbations of respiratory symptoms and on pulmonary function in patients with cystic fibrosis. *N Engl J Med* 1994;**331**:637—42.

8. Patel A, Harrison E, Durward A, et al. Intratracheal recombinant human deoxyribonuclease in acute life-threatening asthma refractory to conventional treatment. *Br J Anaesth* 2000;**84**:505—7.
9. Puterman AS, Weinberg EG. rhDNase in acute asthma. *Pediatr Pulmonol* 1997;**23**:316—7.
10. Greally P. Human recombinant DNase for mucus plugging in status asthmaticus. *Lancet* 1995;**346**:1423—4.
11. Boogaard R, Smit F, Schornagel R, et al. Recombinant human deoxyribonuclease for the treatment of acute asthma in children. *Thorax* 2008;**63**:141—6.
12. Miller MR, Hankinson J, Brusasco V, et al. ATS/ERS task force: standardization of lung function testing. *Eur Respir J* 2005;**26**: 319—38.
13. Silverman RA, Flaster E, Enright PL, et al. FEV_1 performance among patients with acute asthma: results from a multicenter trial. *Chest* 2007;**131**:164—71.
14. Durward A, Forte V, Shemie SD. Resolution of mucus plugging and atelectasis after intratracheal rhDNase therapy in a mechanically ventilated child with refractory status asthmaticus. *Crit Care Med* 2000;**28**:560—2.
15. Hull JHK, Castle N, Knight RK, et al. Nebulised DNase in the treatment of life threatening asthma. *Resuscitation* 2007;**74**: 175—7.
16. McDougall M, Colhurst S, Tibby S, et al. The efficacy of intratracheal rhDNase therapy in the treatment of severe hypocapnoeic respiratory acidosis in ventilated children with status asthmaticus and bronchiolitis. *Crit Care* 2006;**10**:52.
17. Hogg JC. The pathology of asthma. *APMIS* 1997;**105**:735—45.
18. Sidebotham HJ, Roche WR. Asthma deaths; persistent and preventable mortality. *Histopathology* 2003;**43**:105—17.
19. Fahy JV, Steiger DJ, Liu J, et al. Markers of mucus secretion and DNA levels in induced sputum from asthmatic and from healthy subjects. *Am Rev Respir Dis* 1993;**147**:1132—7.